Supporting Information

Tuning LCST with thiol-responsiveness of thermoresponsive copolymers containing pendant disulfides

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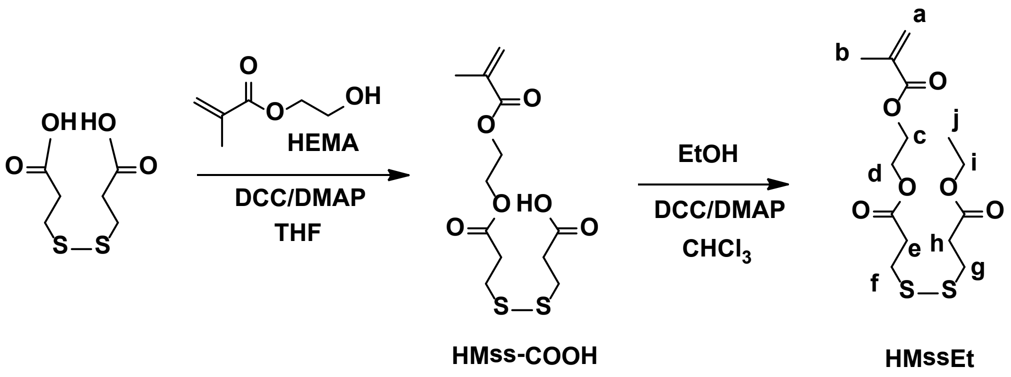
I. Instrumentation and materials

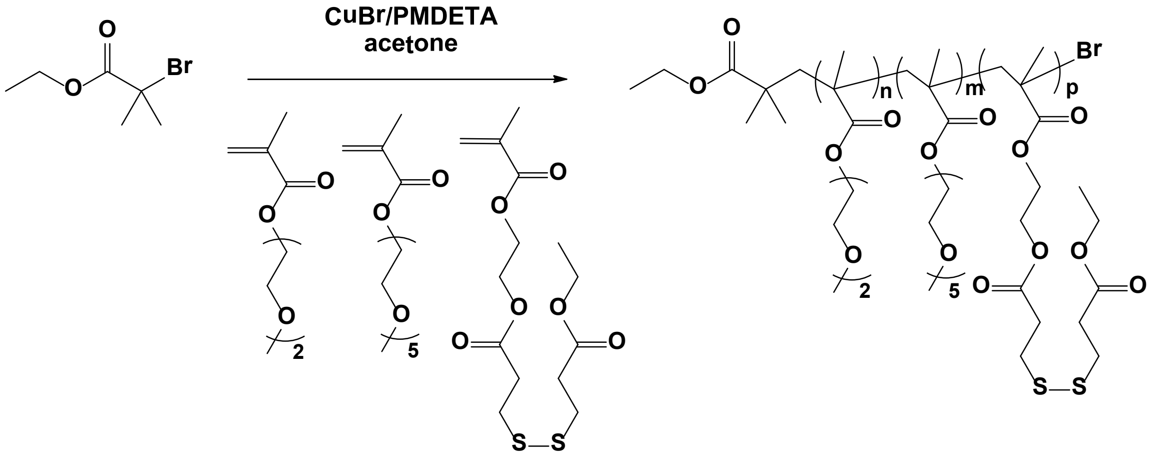
Instrumentation and analyses. ^{1}H-NMR spectra were recorded using a 500 MHz Varian spectrometer. The $CDCl_3$ singlet at 7.26 ppm was selected as the reference standard. Spectral features are tabulated in the following order: chemical shift (ppm); multiplicity (s - singlet, d - doublet, t - triplet, m - complex multiple); number of protons; position of protons. Monomer conversion was determined using ^{1}H NMR. Gel permeation chromatography (GPC) was used to determine the molecular weight and molecular weight distribution of the polymer samples. Samples were prepared by dissolving aliquots of polymer in either THF or DMF containing 0.1% mol LiBr. The dissolved samples were then passed through a column packed with basic alumina to remove any residual copper, before being filtered through a PTFE filter (0.25 µm pore size) to remove any insoluble species. A drop of anisole was added as a flow rate marker. In total two different chromatographs with THF or DMF/0.1% mol LiBr as eluent were used. For samples dissolved in THF, a Viscotek GPC equipped with a VE1122 pump, two PolyAnalytik columns (PAS-103L and 106L) and a refractive index (RI) detector operating at 30 °C was used. The flow rate of THF was set at 1.0 mL min^{-1}. In addition, an Agilent GPC equipped with a 1260 Infinity Isocratic Pump, two Agilent columns (PLgel mixed-D and mixed-C) and a RI detector operating at 50 °C was used for DMF samples. DMF containing 0.1 mol% LiBr was used as eluent at a flow rate of 1 mL min^{-1}. The detectors were calibrated using linear poly(methylmethacrylate) (PMMA) standards from Fluka. The molecular weights of polymer samples were obtained using a calibration calculated from PMMA standards. The molecular weight data for samples taken to investigate polymerization kinetics presented in the supporting information (Figure S1 and S2) were examined using the THF GPC, while molecular weight data for purified polymer samples presented in Table 1 were analysed using the DMF GPC.

Materials. Ethyl α-bromoisobutyrate (EBiB), copper(I) bromide (CuBr, >99.99%), *N*,*N*,*N*′,*N*″,*N*″-pentamethyldiethylenetriamine (PMDETA, >98.0%), and butylamine (99%) from Aldrich and DL-dithiothreitol (DTT, 99%) from Acros Organics were purchased and used as received. Di(ethylene glycol) monomethyl ether methacrylate (DEGMA) with pendant EO units DP = 2, oligo(ethylene oxide) methyl ether methacrylate (OEOMA300) with M = 300 g/mol and pendant EO units DP ≈ 5, and tert-butyl acrylate (tBA) from Aldrich were purified by passing them through a column filled with basic alumina to remove inhibitors. HMssEt methacrylate bearing a pendant disulfide linkage was synthesized in two steps. Briefly, HEMA (10.5 g, 80.9 mmol) reacted with ss-DCOOH (70.0 g, 332.9 mmol) in the presence of DCC (18.0 g, 87.2 mmol) and DMAP (0.88 g, 7.2 mmol) in THF (700 mL), yielding residues containing HMss-COOH. After removal of THF, the intermediate HMss-COOH reacted with EtOH (19 g, 412.4 mmol) in the presence of DCC (20.0 g, 96.9 mmol) and DMAP (0.12 g, 0.98 mmol) dissolved in chloroform (110 mL). Column chromatography was used to purify HMssEt as an oily residue. Yield = 11.9 g (42%). R_f = 0.36 on silica (3/7 ethyl acetate/hexane). ^{1}H-NMR ($CDCl_3$, ppm) 6.12 (s, 1H, $H_2C{=}C$-), 5.59 (s, 1H, $H_2C{=}C$-), 4.35 (s, 4H, -C(O)O$\underline{CH_2}\underline{CH_2}$O(O)C-), 4.15 (q, 2H,-C(O)O$\underline{CH_2}CH_3$), 2,91 (t, 4H, -$\underline{CH_2}$SS$\underline{CH_2}$-), 2.76 (t, 2H, -O(O)C$\underline{CH_2}CH_2$SS-), 2.71 (t, 2H, -SS$CH_2\underline{CH_2}$C(O)O-), 1.94 (s, 3H, $H_2C{=}C(\underline{CH_3})$-), 1.26 (t, 3H, -C(O)O$CH_2\underline{CH_3}$). ^{13}C-NMR ($CDCl_3$, ppm): 171.5, 171.3, 166.9, 135.8, 126.0, 62.4, 62.2, 60.7, 34.1, 33.9, 33.2, 32.9, 18.2, 14.1. Mass calculated for ($C_{14}H_{22}O_6S_2Na^+$): 373.07555. Found: 373.07554.



Scheme S1. Synthesis of of a new methacrylate bearing a pendant disulfide linkage HMssEt.

II. Synthesis of P(OEOMA-co-DEGMA-co-HMssEt) copolymers by ATRP



Scheme S2. Synthesis of well-controlled P(OEOMA-co-HMssEt) copolymers (ssCPs) using atom transfer radical polymerization (ATRP) catalyzed with CuBr/PMDETA in the presence of EBiB initiator in acetone at 47 °C.

A series of thermoresponsive P(OEOMA-co-HMssEt) copolymers containing various amounts of PHMssEt units was synthesized via ATRP catalyzed with CuBr/PMDETA complex in the presence of EBiB in acetone at 47 °C. A typical procedure for ssCP-2 is described as follow; EBiB (39.5 μL, 0.27 mmol), OEOMA300 (2.2 g, 7.3 mmol), DEGMA (3.6 g, 19.1 mmol), HMssEt (0.2 g, 0.56 mmol), PMDETA (23.4 mg, 0.14 mmol), and acetone (7.7 mL) were mixed in a 25 mL Schlenk flask. The resulting mixture was deoxygenated by three freeze-pump-thaw cycles. The reaction flask was filled with nitrogen and then CuBr (19.4 mg, 0.14 mmol) was quickly added to the frozen solution. The flask was closed, evacuated with vacuum and backfilled with nitrogen three times. The mixture was thawed and then the flask was immersed in an oil bath preheated at 47 °C to start polymerization. Aliquots were withdrawn at different time intervals to analyse molecular weight by GPC and conversion by ^{1}H-NMR. Polymerization was stopped by cooling and exposing the reaction mixture to air.

For purification, the as-synthesized polymer solution was passed through a basic alumina column to remove the copper complex, and then solvents were removed by rotary evaporation. The products were precipitated from hexane three times, and then dried in vacuum oven at room temperature for 18 hrs.

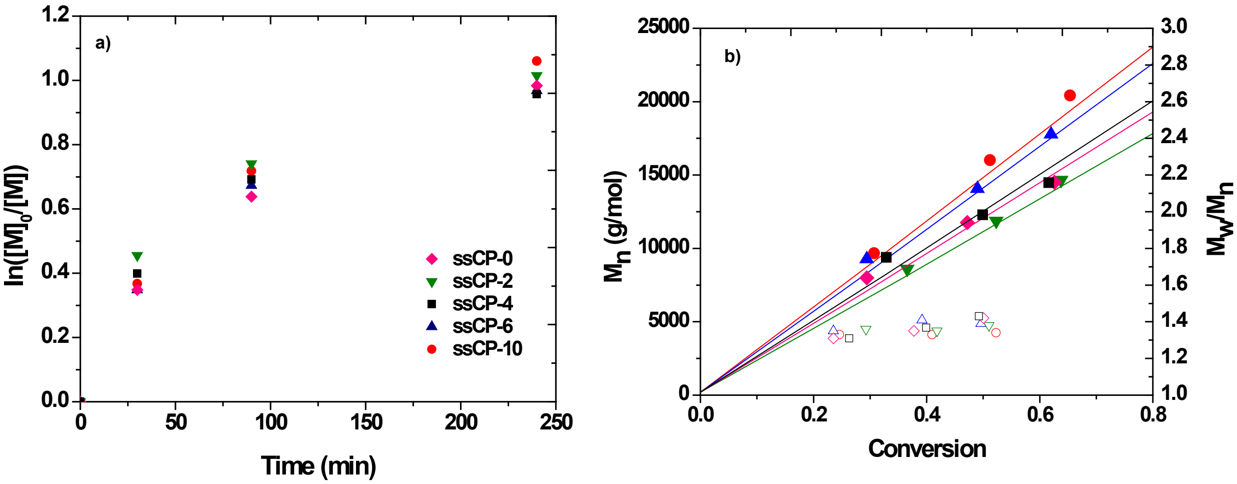


Figure S1. Kinetic plot (a) and evolution of molecular weight and molecular weight distribution over conversion (b) for ATRP of mixtures of OEOMA300 and DEGMA with different amounts of HMssEt in acetone at 47 °C. ATRP conditions: $[\text{Monomers}]_0/[\text{EBiB}]_0/[\text{CuBr}]_0/[\text{PMDETA}]_0 = 120/1/0.5/0.5$ and monomers/acetone = 1 /1 wt/wt. The straight lines in (b) are the theoretically predicted molecular weight over conversion. Molecular weight data were determined using THF GPC.

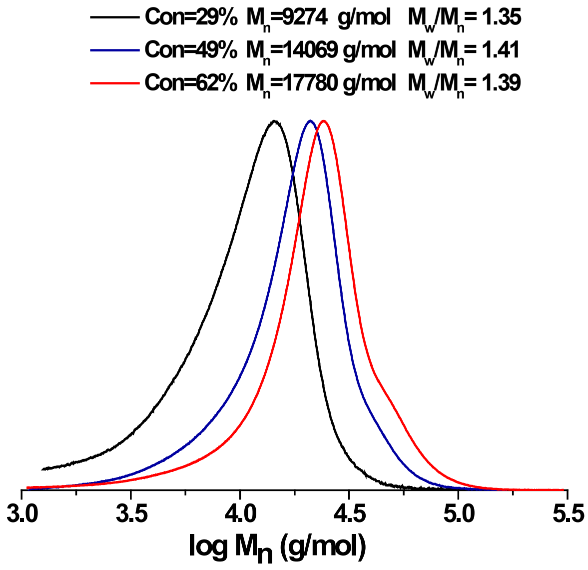


Figure S2. GPC traces of ssCP-6, demonstrating representative evolution of molecular weight over time for well-controlled ATRP experiments. Molecular weight data were determined using THF GPC.

III. Thiol-responsive cleavage of disulfide linkages of copolymers

The purified, dried ssCP was dissolved in DMF (0.5 mL) and mixed with DTT (5 mole equivalents to disulfide linkages in copolymers) under stirring for 18 hrs. The products were precipitated from a mixture of THF/hexane=1/25 v/v three times, and then dried in vacuum for 4 hrs.

IV. Determination of LCST using dynamic light scattering

Light scattering (LS) intensity (count rates) of aqueous copolymer solutions was measured by dynamic light scattering (DLS) at a fixed scattering angle of 173° with a Malvern Instruments Nano S ZEN1600 equipped with a 633 nm He-Ne gas laser. For the measurements, aqueous polymer solutions at a concentration of 0.1 mg/mL were filtered through a 0.45 μm PES filters to remove large aggregates. The temperature was varied from 20 to 85 °C at increments of 1°C and three measurements were made with a total of six scans at each temperature. LCST was determined from the onset of increase in LS intensity.

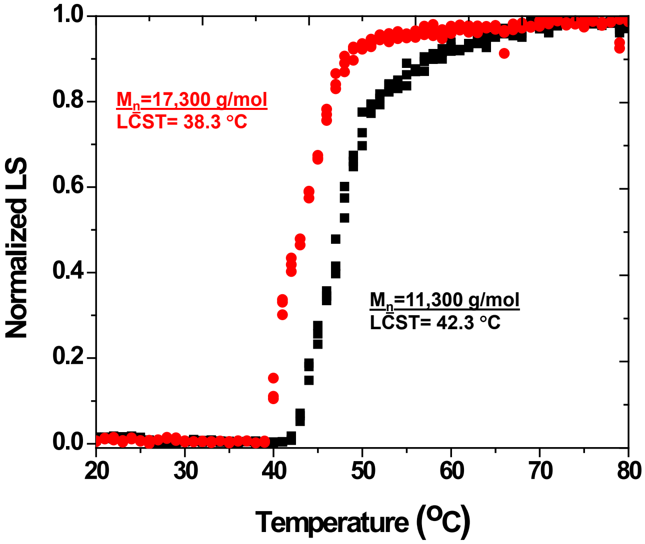


Figure S3. Temperature dependence of normalized light scattering intensity by DLS for 0.1 mg/mL aqueous solutions of ssCP-2 (2% HMssEt) with different molecular weights.

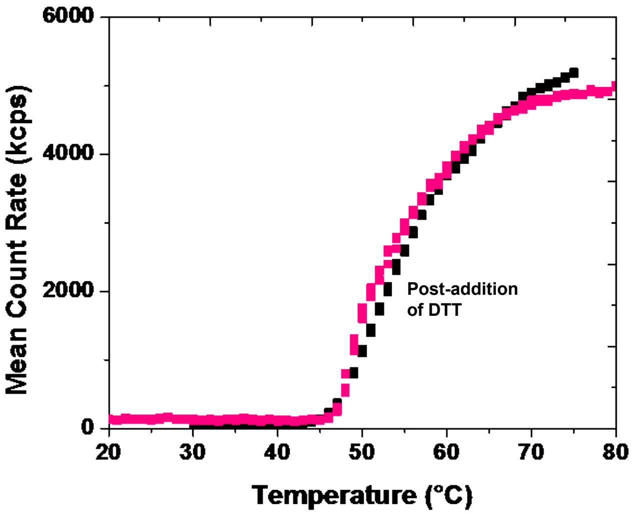


Figure S4. Temperature dependence of light scattering intensity by DLS for 0.1 mg/mL aqueous solutions of shCP-2 (2% HMssEt) upon further addition of DTT to examine the effect of residual DTT on its thermal properties.

V. Modification of pendant thiols by Michael addition to tert-butyl acrylate

Purified shCP-6 (50 mg, 1.6 µmol SH) was dissolved in deuterated chloroform (0.75 mL). The polymer solution was quickly combined with tert-butyl acrylate (0.31 mg, 2.4 µmol) and butylamine (0.18 mg, 2.4 µmol) into an NMR tube and shaken gently. The reaction progress was tracked by ^{1}H-NMR over specified time intervals. To recover the tBA modified polymer, the polymer solution was quickly dried under vacuum, dissolved in methylene chloride and then washed with 5 mL of 0.05 M HCl and NaCl solution. The organic phase was then dried under vacuum at room temperature for 4 hours.

The ^{1}H-NMR spectra of sCP-6 is shown in Figure S5. The conversion of free pendant thiols to sulfides was calculated using the integration ratio of the nine tertiary methyl protons (h) at 1.5 ppm to the methylene protons protons (b, c″, i, j) between 2.4 and 2.9 ppm. Since it was not possible to separate the methylene protons corresponding to free thiol (b, c’) and those belong to sulfide (b, c’’), the integration area of i was assumed to be equal to j, and they were subtracted from the total integration area between 2.7 and 2.9 ppm to give total protons adjacent to free thiols or sulfide bonds. The conversion was then calculated as follows:

$$x_{sulfide} = \frac{\frac{I_h}{9}}{\frac{I_{b,c'',i} - I_j}{4} + \frac{I_h}{9}} \times 100\%$$

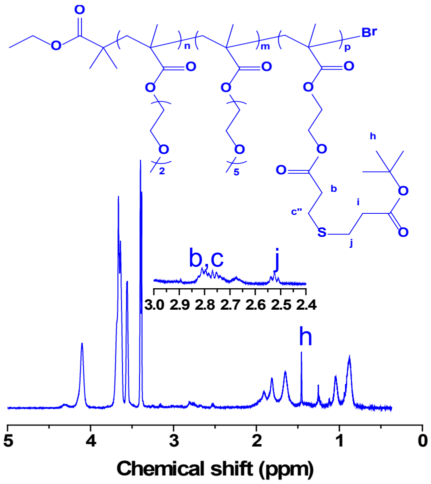


Figure S5. ^{1}H-NMR of sCP-6 with pendant sulfides converted by thiol-ene reactions with tBA.